Effect of Long-Term Hyperbaric Stress on Ammonia Metabolism in Humans

SARA C. GILMAN, W. L. HUNTER, JR., LOREN W. MOONEY, and R. J. BIERSNER

Environmental Physiology and Operational Medicine Departments, Naval Submarine Medical Research Laboratory, Groton, Connecticut 06340

GILMAN, S. C., W. L. HUNTER, JR., L. W. MOONEY, and R. J. BIERSNER. *Effect of long-term hyperbaric stress on ammonia metabolism in humans.* Aviat. Space Environ. Med. 49(9):1093-1094, 1978.

A significant increase in blood urea was found in 11 U.S. Navy divers during 8 d of air saturation hyperbaric exposure. Similar increases in blood urea have been found in animals that convulsed during hyperbaric oxygen exposure. Therefore, it is suggested that careful attention be given to blood ammonia and urea levels in humans during long-term hyperbaric exposure.

THE MAJOR NEURAL MECHANISM involved in stress reactions is the adrenergic nervous system (ANS). Most types of psychophysiological stress produce increased ANS activity (3,4,5,8), accompanied by ammonia liberation in blood and brain (10,12). Stress factors associated with chronic exposure to hyperbaric conditions, e.g. increased levels of oxygen or nitrogen, may also produce increased ANS activity and hyperammonaemia. Elevations in ammonia levels have been found in animals that convulsed during exposure to hyperbaric oxygen (1,2,6). Drugs which are able to chelate ammonia protect against oxygen-induced seizures (1).

In humans, research has shown that amylase secretion, another indicator of ANS activity, is significantly elevated during prolonged hyperbaric exposure (7). If this ANS hyperactivity also results in hyperammonaemia in man, resistance to oxygen-induced seizures could be significantly decreased.

The present research is concerned with the possible hyperammonaemic effects of long-term hyperbaric exposure in humans. Blood urea levels were used as a measure of hyperammonaemia since urea is formed in association with ammonia buffering.

This study was supported by Naval Submarine Medical Research and Development Command Research Work Unit MOO99-PN.001-8013 entitled "Stimulated saturation diving employing nitrogen-oxygen mixtures as the principal breathing media." The opinions and assertions contained herein are the private ones of the writers and are not to be construed as officially reflecting the views of the Navy Department, the Naval Submarine Medical Research Laboratory, or the Naval Service at large.

MATERIALS AND METHODS

Adult male U. S. Navy divers, 11 in groups of three or four each, were exposed to hyperbaric air for 8 d using a 10 × 30 f steel hyperbaric chamber maintained at the Naval Submarine Medical Research Laboratory. The saturation pressure was 2.8 ATA, equivalent to 60 feet of sea water (FSW), with daily, 8-h excursions starting at 10 a.m. to 4.0 ATA, equivalent to 100 FSW. These excursions did not require decompression on return to 60 FSW. A recirculation atmosphere control system was used to regulate P_{O_2} and P_{CO_2} at 20.9% and $< 1\%$, respectively. Temperature and humidity were regulated for diver comfort. Their daily schedule included numerous physiological and behavioral tests commencing at 10 a.m. and ending at 8 p.m. daily. Decompression to the surface required 20 h and commenced at 2 p.m. on the seventh day. Blood samples were obtained daily from each subject during the 3-d pre-dive period, during the 8-d dive period, including decompression, and for the first 3 d post-dive. Samples were obtained daily at 7 a.m. with the subjects in a fasting state. Subjects were given a general diet throughout the study. Alcohol, tea, and coffee intake were restricted. After collection, serum was frozen for subsequent determination of blood urea.

In order to limit statistical analyses to differences occurring across the three test conditions (pre-dive, dive, and post-dive), the mean of the separate collection values for each subject within each of the three test conditions was determined first. An analysis of variance technique applicable to repeated measures (same subjects) was applied to the distributions obtained for the three conditions. Differences between conditions were then tested for significance using t-tests for correlated samples. Levels of significance are $p \leq 0.05$ (two tailed).

RESULTS AND DISCUSSION

Fig. 1 shows the means and S.E.M. for blood urea level (in mg/100 ml) across the three test conditions (pre-dive, dive, and post-dive). The analysis of variance showed that significant differences occurred across these blood urea means ($F = 14.58$; $df = 2,3$; $p < 0.01$). Sub-

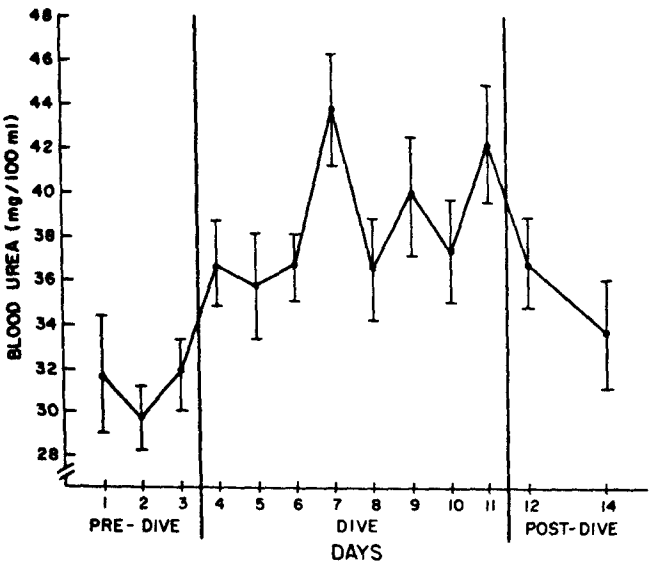

Fig. 1. Mean and S.E.M. for blood urea levels of the 11 divers during the 8 d of hyperbaric exposure.

sequent t-tests demonstrated that the dive mean was significantly higher than the pre-dive mean ($t = 5.95$; $df = 10$; $p<0.01$). Mean blood urea levels during the post-dive period, although declining, did not differ significantly from the dive means ($t = 1.73$; $df = 10$; $p>0.05$) but were significantly higher than pre-dive means ($t = 3.66$; $df = 10$; $p<0.01$). These results indicate that blood urea was significantly elevated during the dive and, although declining, did not reach pre-dive levels during the three post-dive days.

The increase in blood urea in these subjects during long-term hyperbaric exposure is noteworthy in view of previous animal studies showing that significant elevations in blood ammonia were found in animals that convulsed during hyperbaric oxygen exposure (6). More recently, Banister *et al.* (1,2) reported significant elevations in blood ammonia and urea in rats exposed to oxygen at 6 ATA. The largest increase occurred following exposure periods of sufficient duration to produce convulsions. A 300% increase in blood ammonia and a 69% increase in blood urea over control values was found in the convulsed animals. Pre-treatment with lithium, which reduced the blood ammonia level, prevented the convulsions. Little information is available about the involvement of ammonia in the development of oxygen toxicity in humans. This study shows that an average increase of 23.4% in blood urea occurred over the 8 d of hyperbaric exposure. The increase is progressive, reaching a maximum of over 37% by the last day of exposure. However, no signs or symptoms of CNS oxygen toxicity were seen. Despite the absence of such symptoms, these findings, when coupled with those from the animal studies referred to previously, suggest that careful attention should be given to changes in blood ammonia and urea in humans during long-term hyperbaric exposure.

However, monitoring blood levels of ammonia and urea during hyperbaric exposure entails many operational and technical problems, e.g. sending a phlebotomist to depth, blood loss, time-consuming chemical assay, etc. A speedy noninvasive measure requiring small sample volumes would be preferred. The analysis of parotid gland excretion of urea using the electrode method of Renfro and Patel (11) may provide such a method.

Kopstein and Wrong (9) have shown that the urea concentration in saliva from the parotid duct is positively correlated with plasma urea concentration. In fact, parotid urea has an average urea concentration of 86% of the plasma concentration. Therefore, subsequent studies will investigate parotid urea excretion during hyperbaric exposure in relation to the plasma concentration.

ACKNOWLEDGMENT

The authors wish to thank Ms. Pauline K. Sokolski for her valuable assistance in the preparation of this manuscript. We also wish to thank Lakeville Medical Laboratories, New Hyde Park, NY, for technical assistance and support.

REFERENCES

1. Banister, E. W., N. M. Bhakthan, and A. K. Singh. 1976. Lithium protection against oxygen toxicity in rats: Ammonia and amino acid metabolism. *J. Physiol.* 260:587-596.
2. Banister, E. W., A. J. Davidson, N. M. Bhakthan, and C. Asmudson. 1973. Biochemical effects of oxygen at high pressure in rats. *Can. J. Physiol. Pharmacol.* 51:673-678.
3. Bloom, G., U. S. von Euler, and M. Frankenhaeuser. 1963. Catecholamine excretion and personality traits in paratroop trainees. *Acta Physiol. Scand.* 58:77-89.
4. Frankenhaeuser, M. 1975. Experimental approaches to the study of catecholamines and emotion. *In*: L. Levi (Ed.), Emotions: Their parameters and measurement. New York: Raven Press.
5. Frankenhaeuser, M., and P. Patkai. 1965. Interindividual differences in catecholamine excretion during stress. *Scand. J. Psychol.* 6:117-123.
6. Gershenovitch, Z. S., and A. A. Krichevskaya. 1960. The protective role of arginine in oxygen poisoning. *Biokhimiya* 25:790-795.
7. Gilman, S. C., D. A. Miller, R. D. Thornton, G. J. Fischer, and R. J. Biersner. 1978. Hyperactivity of the human parotid gland during chronic hyperbaric exposure. Abst. Proc. Undersea Med. Soc., April.
8. Goodall, C. M., M. McCally, and D. E. Graveline. 1964. Urinary adrenaline and noradrenaline response to simulated weightless state. *Am. J. Physiol.* 206:431-436.
9. Kopstein, J., and O. M. Wrong. 1977. The origin and fate of salivary urea and ammonia in man. *Clin. Sci. Molec. Med.* 52:9-17.
10. Quastel, J. H. 1974. Amino acids and the brain. *Biochem. Soc. Trans.* 2:766-780.
11. Renfro, J. L. and Y. Patel. 1974. Urea analysis with the ammonia electrode. *J. Appl. Physiol.* 37:756-757.
12. Tsukada, Y. 1971. Ammonia metabolism. *In*: A. Lajtha (Ed.), Handbook of Neurochemistry V. New York: Plenum.